

DATE:

July 7, 1984

REF:

Y61

W 25 83 X 2639 5:5:10 12:15 PM - ca 2:30 PM

(As many pairs became stuck there is no advantage to using excess of non-motile parent!)

A	1	4	cell	drop	lac
	2	2	28	28	
	3	1	1	1	
A	4	X	1B	1	
B	1		00.	28	
	2		1	0	
	3		1	1	
B	4		0.	28	
	5		1	1	
C	1		00.	28	
	2		1	1	
	3		1	1	
C	4		00	28	
	5		00	1	
	6		00	1	
D	1		00	1	
	2		0	1	
	3		0	1	
D	4		00	4/28	
	5		0	1	
	6		0	1	
E	1		1.	1	
	2		1.	1	
E	4		0000	28	
	5		1	1	
F	1		00	28	
	2		1.	1	
	3		1.	1	
F	4		0.	28	
	5		1	1	
	6		1	1	
G	1		00	28	
	2		1	1	
	3		1	1	
G	4		00	28	
	5		1	1	
	6		1	1	
H	1		0000	28	
	2		0 lost?	0	
H	4		1	28	
	5		00	1	
	6		00	1	

1 - lac -
28 - lac +
except E5.
lac

prob not covered

very hard

00 2?

♂ x ♀

162

July 8, 1954

~~105~~ - 105 - 250 +

(Kiekenby demerits.)

♂ x ♀
drop 28

1:10
later high incidence of pairs.

Group	Cell	♂	♀	1:10
A1	0	0	1	+
A2	0	0	0	+
A3	0	0	0	+
A4	0	0	0	+
A5	0	0	0	+
A6	0	0	0	+
B1	0	0	0	-
B2	0	0	0	-
B3	0	0	0	+
B4	0	0	0	+
B5	0	0	0	+
B6	0	0	0	+
C1	0	0	0	+
C2	0	0	0	+
C3	0	0	0	+
C4	0	0	0	+
C5	0	0	0	+
D1	0	0	0	+
D2	0	0	0	+
D3	0	0	0	+
D4	0	0	0	+
D5	0	0	0	+
D6	0	0	0	+
E1	0	0	0	+
E2	0	0	0	+
E3	0	0	0	+
E4	0	0	0	+
E5	0	0	0	+
E6	0	0	0	+
F1	0	0	0	-
F2	0	0	0	-
F3	0	0	0	-
G1	0	0	0	+
G2	0	0	0	+
G3	0	0	0	+
G4	0	0	0	+
G5	0	0	0	+
H1	0	0	0	+
H2	0	0	0	+
H3	0	0	0	+
H4	0	0	0	+
H5	0	0	0	+
H6	0	0	0	+

but see scores after 1179
score dubious
not counted

not spec. looked for
presumably absent!
(light body?)

Many inviable. Complete marked
No (2)!

F "Kump"

1178
(1177)

July 10, 1954.

164

W2640: W2639 1:50. $105 - 320$ pairs ~~with~~ infrequent
 either paired at ca 3^{30} while isolated.

		deep	#	interest
A1	0	28	39	
2	1	0		
3	1	1		P F+ ✓
A4	00	28	40	
5	1	1	2	P
6	1	1	3	P P?
B1	X	1	4	P
2	1	1	5	P
B4		1	6	
3		0	7	IP
5		1	8	
6		1		

15 del. random. 0 F+
 7 del pairs 2 F+

sup C

1	0	28	41	
2	0	1	109	P
4	00	0		
M. 5	1	0		
6	1	1	10	P'

D2	X	0		
3		1	12	X

D4-6, E4-6, F1-6, G1-6 are random isolates of motiles (at end)

H 1			33	} IP F+ }
2	all motile, not pair.		34	
3			35	

H 4	0	1-28	36	X	(28+1).
5	0	1	37	P	F+
6	0	1	38	P	F+

E 1	0	28	42	
2	1	1	16	P
3	1	1	17	P

and 37, 38
 1-18 should be checked as pair if progeny
 39-42 are pair if P+
 6-8 and 33-55 maybe illegitimate pairs
 to E 17L (paired to 1 and progeny P11)

DATE: July 12, 1954

REF: [158] [165]

X 2 - 2+ hour intervals as in pre-pair experiments.
crosses might have been confused?

Note fairly numerous colonies type 28 lact and -

	1	2	3	4	5	6	7	8	9	10
	lac type (18)									
10	E4 -	A1 -	B5 -	D6 +	G4 -					
	E1 -	2 -	6 -	E2 -	5 -					
	E2 -	3 -	C1 +	3 -	6 -					
	E2 -	4 -	2 -	4 +	H2 -					
	H1 +	5 -	3 -	5 -	3 -					
		6 +	5 -	F1 -	4 -					
		B1 -	D1 -	4 -	5 -					
		2 -	2 -	5 -	6 -					
		3 -	3 -	6 -	6 -					
		4 -	4 +	G3 -						
20										
	A1 -	A3 -	D4 -	F5 -						
	2 -	5 -	5 -	G1 +						
	4 -	6 -	6 -	2 +						
	B5 -	B1 +	E1 -	4 +						
	C1 -	2 -	3 -	6 -						
	C2 -	4 -	4 -	H1 -						
	C3 -	6 -	5 -	2 +						
	C4 -	C4 -	F1 -	6 -						
	D1 -	5 -	2 -	6 -						
	D2 -	D1 -	4 +							
	D3 -									
	D4 -									
	D5 -									
	D6 -									
	D7 -									
	D8 -									
	D9 -									
	D10 -									
	D11 -									
	D12 -									
	D13 -									
	D14 -									
	D15 -									
	D16 -									
	D17 -									
	D18 -									
	D19 -									
	D20 -									
	D21 -									
	D22 -									
	D23 -									
	D24 -									
	D25 -									
	D26 -									
	D27 -									
	D28 -									
	D29 -									
	D30 -									
	D31 -									
	D32 -									
	D33 -									
	D34 -									
	D35 -									
	D36 -									
	D37 -									
	D38 -									
	D39 -									
	D40 -									
	D41 -									
	D42 -									
	D43 -									
	D44 -									
	D45 -									
	D46 -									
	D47 -									
	D48 -									
	D49 -									
	D50 -									

O recorded as 28 days

[158]

[165]

10/8
what is this
wpt?
Where are
subjects.
presumably occurrence
of ⊕ among mycamp
& isolates.

all + should be checked for virulence

Sept. 20, 1954

At my request for "the aerogenes strain used in the Baskett-Hinshelwood expts. (FRS 139:58-73, 1951) received a culture labelled simply "Aerogenes aerobacter" 19.7.54. This is stated to have a lag of about 5-6 days in synthetic-arabinose media.

Initially it was streaked on EM-B-L-arabinose and found positive.

Alek Bernstein received culture and stored it as W-2654. For first experiments, slant from single colony on L-arabinose was used. Subsequently, used slant directly from Hinshelwood's vial.

9/20. PM. Inoculate D(m) (citrate!) and D(O) for inocula. Latter grew well in 24h; former shows slight initial growth.

p21: From D(O) above, streak out EM-B-Darabinose (Dar) and inoculate: (.1ml / 10)

	A22	P22	A23	P24	P25
D(m)	±	✓	+	+	
D(O)	+++	✓	✓		
D(m, Dar)	±	± ... + ?	+	+	
(to avoid citrate T(m) until D(A)- T(glu) D(m) s/citrate T(Dar) mix is made up)	÷		±	±	
	++		±	+	
	÷		±	+	

÷ is faint turbidity, scarcely more than medium. Should try smaller.

- A1. EM-B-Dar plate all negative. (faint pink beg. afternoon)
- A2. P22. Streak out from D(Dar) above which shows some growth progress? all negative. No papillae seen. (Blac)
- B1. Restreak original W2654 for single colony for initiation. Prepare current slant and D(O) maximum tube from this.
- P24. Streak out ① from T(Dar) ② ^{sole} papilla on 1 colony of A2 ③ ~~EM-B~~
- C. ^{sole} papilla on 1 colony of A1. to EM-B Dar. In future expts., minimal medium D(Ar) is based on salts & citrate.
- D. Inoc fresh D(O) culture from B1 to D[±]Ar for near selection. 1 = .1ml 2 = .01ml
- E. ~~TAr. to D[±]Ar, D(O)~~ P25: +, +++. Revue. TAr is +++ (EO) N26 E16

P25 N26 A27 P28

D2: D(0) +++ ✓
 D(1-) - ✓ ⊕
 D(A₁) ≠ ± ✓ ✓
 D1: D(0) +++, +++, ✓
 D(1-) ± ± (P₂₈?) ⊕
 D(A₂) ± ± ✓ ✓ ✓

P25 and
 C1-2-3 no + but
 some difference in shading
 more papillae now seen on
 A1-A2
C56 ~~C7~~

P25

F1 = E0 on EMS Pan.

N26: C1 } mostly slow +
 (48h.) 2 }
 3 } mostly -, 1 colony "+"
 (24h.) F1 } mostly (heterogeneous) weak +
 C5 }
 C6 }
 C7 } distinctly two colony types - and +
 no strong +

New plantings N26: E1 is still slower or almost than original.
 Wait on the D series for definitive series; meanwhile sub. E series to
 look for fast arachnoid. Note: C3 is a + from first stage papilla
 of A1. Replete C3, C7 +/- and ✓ on D(0), D(A₁).

EMS	A27 D(0)	D(A ₁)	P28	A0, D(A ₁)
A27: C3A -	+++	-	-	E1 A +++
C3B ± centus	+++	±	+	
C7A + and +	+++	+	+++	
C7B -	+++	-	-	

h. 1ml/10
 ha. P28 C7A'

DATE:

REF:

NB. Since yesterday, 1 tube of D1 has begun to grow.
(Other D1 and D2 still negative).

C7A⁺ seems now to be as rapid ~~as~~ on glucose and arabinose.
On EMBOys, still weak.

Plan (1) Restreak DIA, C7A on EMB, DAr and moi. 1g, D(Ar) for comparison.

P3-P4 (24 hours) — C7A⁺ has formed colonies, other two are similar, seripoints. DIA has scarcely begun.

P4: 30 In 24 hours, C7A⁺ has grown optically (lag = glucose)

Plate moi 9/30, D(Ar). shows C7A⁺ forming good size colonies (two sizes). W2654 forms numerous seripoints & definite stimulation from Ar⁺.

Conclusions (1). EMB probably not a good indicator for this problem. However papillae on EMB suggest a non-homogeneous response (contra Hinshelwood). (2) possibility of trying indirect selection on DAr.

Take C7A⁺ single colony to USA as 1181A.

P4: Note original 9/30 DAr plate shows no papillation yet for W2654 stock.

P6 on D(Ar), DIA shows about 1% dense colonies

P7 puts + and - to D(0) for check.

P6: D2A is now ++.

P7: D2A ++
D1A (++)
D1B now +.

strains G1-3

not adapted?

P11: 1 all v.s.
2 mostly +, few v.s.
3 " " }

still heterozygous

P17) D1+
D1- colony to D(6).
w2604

H	3	+ by P11	A12	A14
	2	-	-	-
	1	-	-	-

∴ these are distinct.

more D(6) all +++ in vph.	D(11) all - still vph
---------------------------------	--------------------------

↓ P14

1. + and - colonies. ○ ○ about =
2. ++ and a few - ○ ○ any +?
3. " " ○ ○

∴ heterogeneity is still obvious.

None of these grows nearly as well as an D(6)er. What was Dean's finding? - Write H?

10/15/54

Embryonics

① Initial strain, H_2S^- grows v. slowly on agar, gives v. small flat colonies which appear to deac. slowly as original.

Mutants not observed on D(Ar) (except possibly after 3-4 weeks) but heavy media have not been tested

② In D(Ar) liquid, H_2S^- grows initially to ca 10^{10} / ml. Then stationary turbidity for 4-5 days, then slow growth.

③ Platings of these first cultures show mixture of Ara- and Ara \pm (denser, faster colonies) on D(Ar) agar. Only 1 trial for evolution of colonies with subsequent lag time.

④ These cultures still have long lags on H_2S^- , but successive transfers are gradually shorter. After 4-5 transfers, fully adapted cultures are found.

⑤ No critical experiments on deadaptation except from 1st stage.

⑥ Platings on D(Ar) and EMBA for at various stages suggest several mutational steps.

A. Cultures adapting in D(Ar) liquid are not homogeneous.

B. \therefore Not proven whether induced or selected.

C. Needed: ① Platings of dense suspensions & hope of identifying

the first step mutants ⊙ lac^+ / lac^- markers
for confirming heterogeneity of response.

Interactions in phenocopy "F"

1182

DATE: Sept. 22, 1954.

REF:

	1	2	3	4	5	6	7	8	9	10
	i' Luca.									
	P21 inoc 10ml Penassay									
	i W6, W1177, W2207, W1305, W2437									
	M-F+ TL-F- M-F+ MTL-F+ MTL-F-									
	.5ml per.									
10	10:20 A22 inoc 10ml Penassay i and s aeration.									
	Plan: set up all combinations (A,B,CD)(1,2,3,4) and wash, cross mixtures to W1177 (aer.) to test interactions. Plate comparable aliquots on D(0) agar									
	W1305	-	A							
	"	+	B							
	W2437	-	C							
	"	+	D							
	W6	-	1							
	"	+	2							
	W2207	-	3							
	"	+	4							
20	3:45 mix 1.5ml each culture + 7ml Penassay. - 4:45 spin down									
	5:30 Resuspend in 1ml water each. 20ml culture (incubate & aerate) W1177 to 5 5ml.									
	Plate $\frac{1}{2}$ W1177 each plate. 1ml others									
30		1		2		3		4		
		W-6 -		W-6 +		W2207 -		W2207 +		
A	F+	W1305	22	17, 17		30, 84		153,		
B		" Aer.	3	7, 2		2, 5		18,		
C	F-	W2437	60	15, 6		0 0		0		
D		" Aer.	29	10, 10		0, 0		0		
E		Penassay.	131	22 22, 25		0, 0		1, 0		
40										
50										
	E2-									
	E3- 0	A x 0								
	E1- 0	C x 0								
	C- 0									
	A- 0									

This is not meaningful continuation by Luca.

Key Submissions.

1182
conclusions

- ① Recovery of acrated cells in Penassay.
- ② Effect of 1 hour acration on unacrated cells.
- ③ Effect of acrated ~~W~~ 1305 on ~~W~~ unacrated.

Comparisons

- ① E2: either acration was ineffective or recovery in Penassay.
- ② 4 series: a. W1305 converted. W1305 acrated still converted but qualified by ①
- 3 series: ~~W~~ W2207 acrated also converted, less effectively ~~by~~ acrated W1305.
- ③ 2 series: see ①. However B2/E2 suggests that 1305 F⁺ an. inhibits recovery.
- ④ 1 series of A1/B1/E1: W1305 acrated may ferment unacrated to W6 unacrated.

$$W1305 F^a \not\rightarrow W6 F^+$$

$$F^+ \not\rightarrow F^a$$

DATE: Sept. 27, 1954.

REF: 173-174

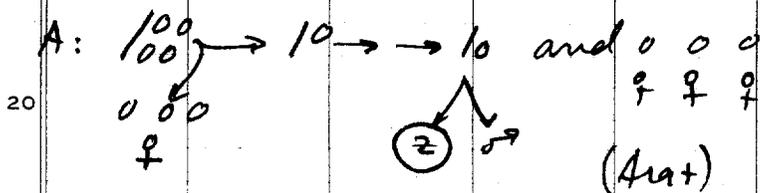
Obj: Begin to look for pedigrees on the ♀ side. Do not try to obtain ♂
pedigrees but record viability and isolate pools.

9:50 AM. Mix overnight cultures 1 ml ♀ : 1 ml ♂ : 10 ml necessary 37°.

9/28 (PM) Plate drops See protocols for pick schedule and pedigree details.

10 Plate to EMS lac. Score P29 and A30.

A30: all parental on lac except A2 and E16. Pending further tests, the
save results may be summarized:

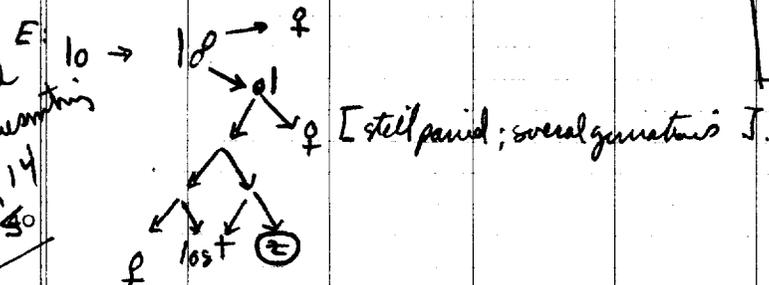


HC!
= still segregating. Might represent $n=0$, $n=2$ or $n=4$.

B: 10 all died

C: $\begin{matrix} 10 \\ \swarrow \searrow \\ \text{♂} \ \text{♀} \end{matrix}$ parental. [1/4 ♀ died]

D: $\begin{matrix} 10 \\ \swarrow \searrow \\ \text{♂} \ \text{♀} \end{matrix}$ died.



F: 10 parental. Note: [3/4 ♀ died] [several generations]

G: $\begin{matrix} 01 \\ \swarrow \searrow \\ \text{♂} \ \text{♀} \end{matrix}$ died.

H: $\begin{matrix} 01 \\ \swarrow \searrow \\ \text{♂} \ \text{♀} \end{matrix}$ (only "1/8" survived).
 parental.

all other isolates concordant (♀ par.) on
Lac, Lac⁺, Mal, Mtl, Xyl, Ara, Gal.
~~(not tested)~~

∴ $n \times 2$, still segregating
[Note 2/4 sibs lost].

∴ 2② from maximum of
5 possibilities. Considerable
mortality in the latter.

save E16 and pools representative E11, B, 14 D6, E50

try H x H for heterozygosity

W2344?

Summary on pairs.

9/23/54

*presumably
no losses on
♀*

NOV 28 1955
Total pairs 11184
4474 ♂ 6710 ♀

DATE	page		Pool.	Intact	c ⁵⁹	Zygotes.	7	8	in 299.	with kid
E. apt.		128	129	4	1	4	2	✓	1	0
		55A	130	4	3	4	4	✓	3	3
		56	131	15	5	9	7	✓	11	5
		[57	11177X	7	2	3	0		5	0
10		58	sip	10 5	8 2	8 4	0	✓	8	0
		59		14	4	8	3	✓	12	2+1?
		60		5	2	3	1	✓	5	1
		61	142	17	-	8	2	✓	14	1
20		74	160	16	10	12	1			
		76	161	14	9	11	1			
		77	162	15	5	10	0	✓		
		66	153	8	8	8	2	✓		
		62	137	16	2	6	1	✓	8	1
30		63	150	12	6	7	3	✓	11	3
		64	151	15	5	12	3	✓	14	3
		65	152	18	8	9	4	✓	12	4
Σ		counted	166 158 166	62	98 ¹⁰¹	34	101 ⁹³	34 ³²		
40		68A	140	4	4	4	0			
		73A	158	13	2	6	1			
		B	159	16	9	13	0			
		74	160	16	10	12	1			
		76	161	14	9	11	1			
50				63	34	46	3			
				229	96	144	37			

do not label empty pairs

*see further for
107 details 25*

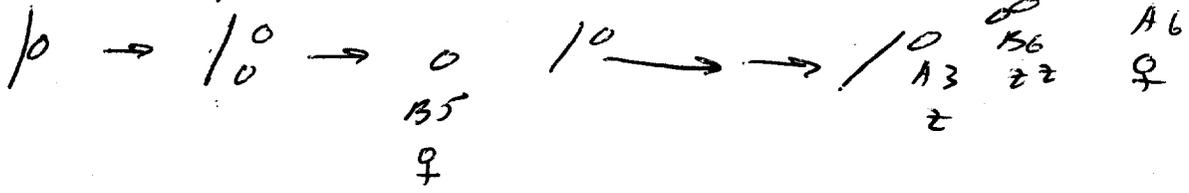
*2 dead
pairs*

()

for

19.211164	151	B3	+		Lact+
20.28		C6	+		Lact+
21.21		D3	+		4 types: [Ar, Lac] ⁺ [Mal, MH, S] ^{+S} -R χ^2
22.301165	152	A3	+		} Lact } Lact
31	A	B6			
32	B				v.c.
23.33		D2	+		} A: Lact (Mal) } B: 4 types [Lac ⁺] [Mal, S] ^{-R} MH ^{-?}
34					
24.36		E3	+		Lact
25.36		H3	+		Lact
26.1166	153	C4	+		Lact
27.38		C3	+		} Lact } Lact
30		4			
28.191159	156		-	pair to C1 comp	Lact
29.91156		C1	+		Lact
30.10131		C3	-		Lact
31.11		D1	+		Lact
32.12		F3	-		Lact
33.13		G5	+		Lact

1165A3-B6 (Hyp. restructure)

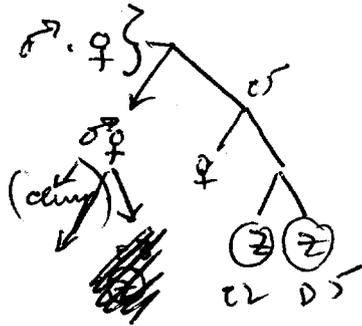


1161G1 = ρ^0/ρ

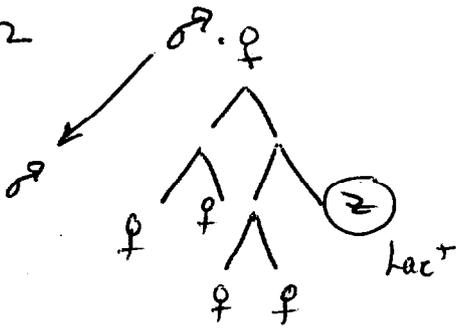
1153B5

(ρ^0/ρ)
(ρ^0/ρ)

1159-22



1160-2



but > 34 zygotes!

Notes on summary:

- ① all zygotes need to be tested for segregation of V_1 , As .
(cf. notes on colony segregation).
- ② of 33 zygotes, 5⁺ survived in 26. Probably as high as controls.
- ③ Pairs after Linsen analysable in 8, 7 are completed. (Exc. 5665)
- ④ Pedigrees 2 or more generations in 7. all still segregating!

Following in z/total:	11912	calc.	2/8	(sibs)
	1160-2	"	1/4	
	61-24		1/4	(1x)
	65-H3, B6.		3/8	(sibs)
	65D2		2/8	
	H3		1/4	
	27C3		4/8	

⑤ Review distribution of Mal. (#16 uncutani). 3 cases of Mal +
all segregating! B5 should be examined for S^+ also say
2 cases of S^+ and both show 4 phenotypes!
(Recall test for recurrent recombination).

⑥ of 34 zygotes. 28 are lac⁺...

DATE:

REF:

1	2	3	4	5	6	7	8	9	10
Among lac^+ :		Ara^+	Ara^-						
	$V_1 R$	(12) \pm	5	(17)					
	$V_1 S$	3	20 \pm	23					
		(15)	25	40					

10 Ara and V_1 are closely linked to each other. Are they linked to lac ? The parents in coupling with lac^+ are circled. There is a definite excess of recombinants, possibly significant (no!) However, incidence of coupling may be exaggerated by admixture.

20 ~~Table~~ Table above is uncorrected for a few sib zygotes. Note especially 1165 B6A, B (31, 32).
30

30

40

50

DATE: Sept. 30, 1954

REF: 175-177

Second run. overnight cultures, 1♀ : 1♂ : 7ml broth 32°
 10²⁰ - 1140 to set up 8 pairs isolated initially, 12:20-12:40 PM.

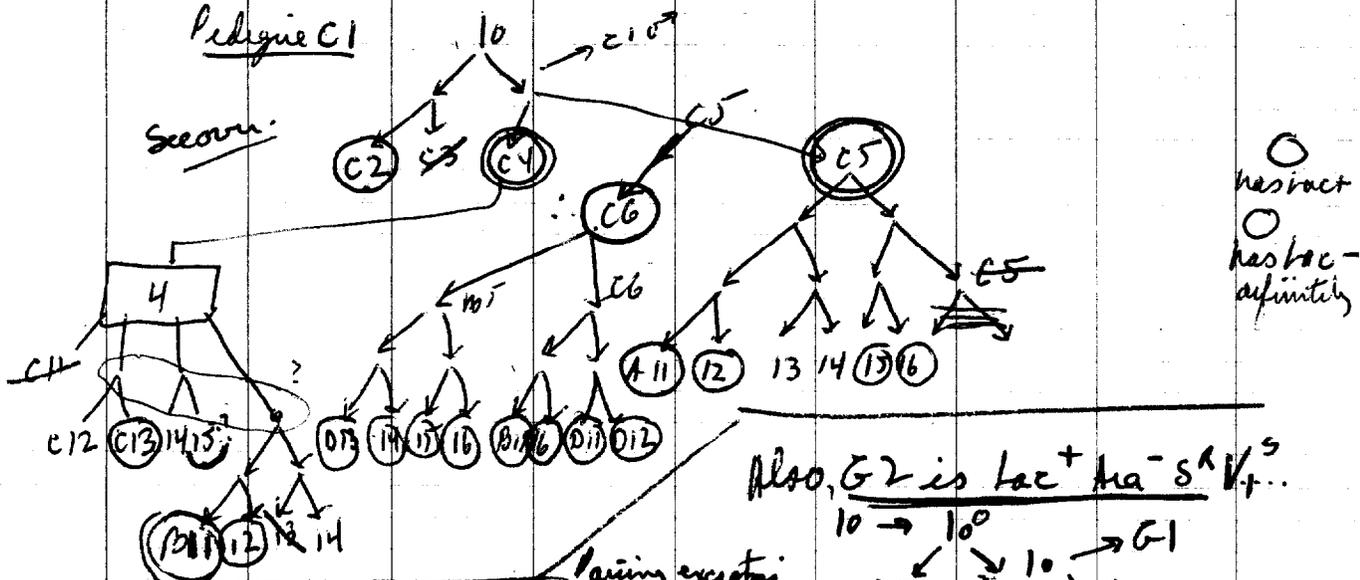
E1 was initially suspicious and proved illegitimate. No viable O from A1; (Secorn)
 paired exconjugant from E1 also inviable. Other pedigrees to 4-6 generations.

See protocols for picking schedule on rows I-VI (lac - or lac+ parents)

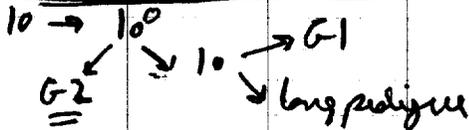
	1	2	3	4	5	6	7	8	9	10
III	A 11	+	+	-	-	-	++	R +	+	V ₁
	12	+	+	-	-	-	++	R +	+	V ₁ R
	13	-	-	-	-	-	++	R -	-	V ₁ S
	14	-	-	-	-	-	++	R -	-	V ₁ S
	15	+	+	-	-	-	++	R +	+	V ₁ R
	16	+	+	-	-	-	++	R +	+	V ₁ R
	B 11	+ -	- , +	-	-	-	++	R +	- +	+ V ₁ R - V ₁ S
	12	+	+	-	-	-	++	R +	- +	+ V ₁ R - V ₁ S
	14	-	-	-	-	-	++	R -	-	V ₁ S
	15	+	+	-	-	-	++	R +	+	V ₁ S
	16	+	+	-	-	-	++	R +	+	V ₁ S
IV	C 12	-	-	-	-	-	++	R -	-	V ₁ S + V ₁ R
	13	+	+	-	-	-	++	R +	+	- V ₁ S
	14	-	-	-	-	-	++	R -	-	V ₁ S + V ₁ R
	15	- +	- , +	-	-	-	++	R +	+	- V ₁ S
	D 11	+	+	-	-	-	++	R +	+	} V ₁ R
	12	+	+	-	-	-	++	R +	+	
	13	+	+	-	-	-	++	R +	+	
	14	+	+	-	-	-	++	R +	+	
	15	+	+	-	-	-	++	R +	+	
	16	+	+	-	-	-	++	R +	+	

Pedigree C1

Secorn:



Also, G2 is lac⁺ tra⁻ S^R V⁺



See G1, G2, pool = 63456 H11-16

Pairing exceptions

O has tract
 O has lac-
 definitely

except for this pedigree, other isolates are
enriched in the regions indicated

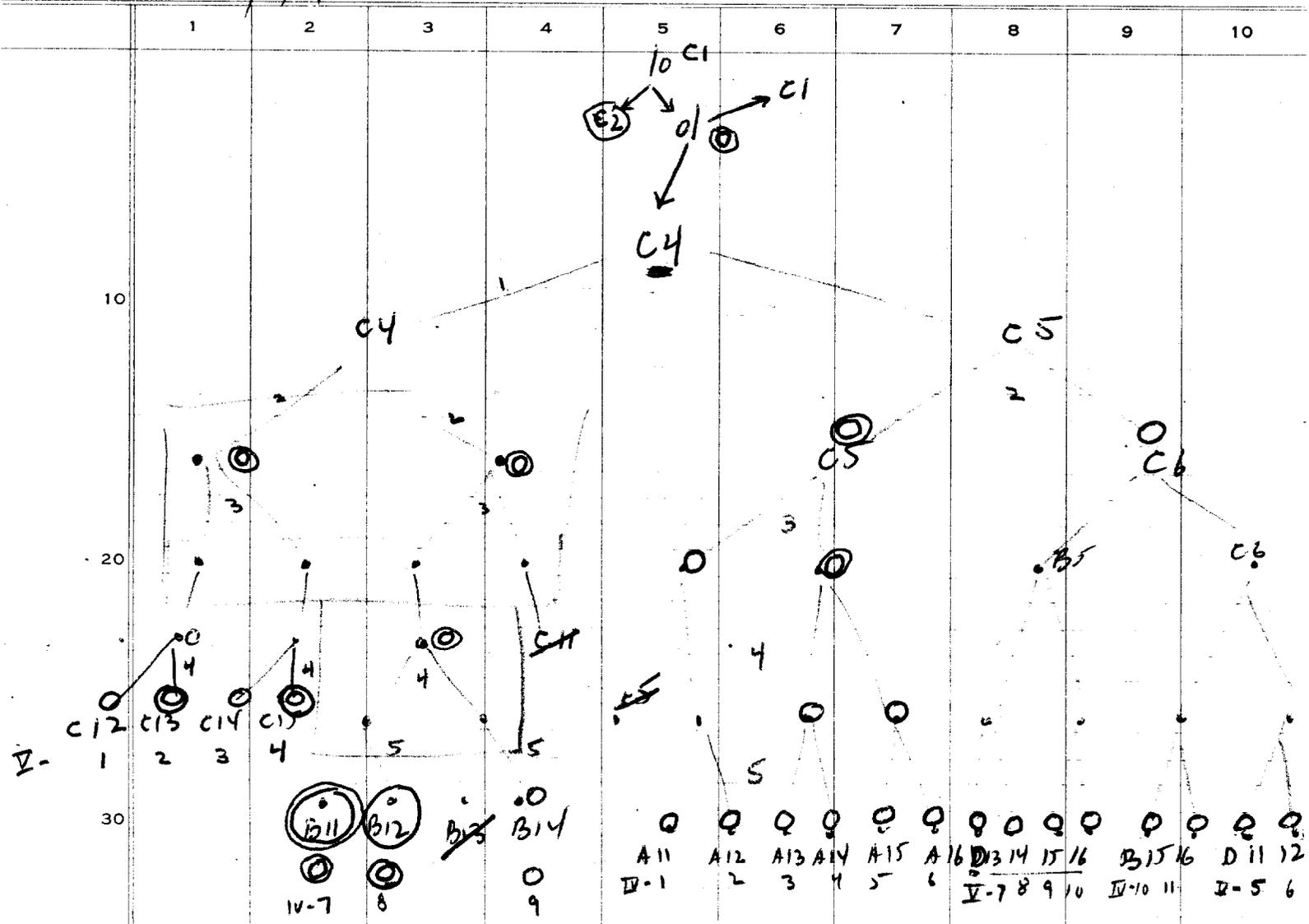
In all likelihood, a single recombinant is
represented, though of the most common type:

$$\left(\frac{\text{Lac}^+ \text{Ara}^+ \text{V}_1^R}{\text{Lac}^- \text{Ara}^- \text{V}_1^S \dots} \right)$$

(absence of other recombinants argues against
double misis)

DATE: 10/2/24

REF:



∴ B11 shows ~~one~~ two lines still segregating after the 5th generation, while the c6 clone seems to have segregated at the 2d. A13-14 / A15-16 probably at the 4th. Pedigree generally should probably be carried to 4 generations.

○ Recamb
 □ ♀ parent

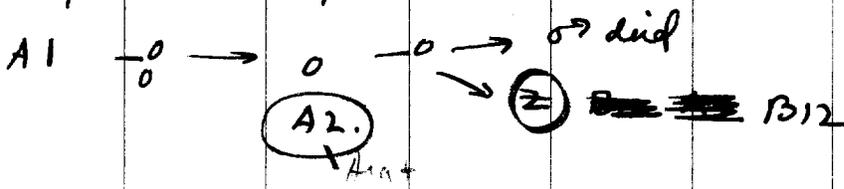
Note: both c4, c5 are 2y, the time of fertilization?

	1186			
	→	♀	Ⓢ	
A1	x	✓	✓	lact... ; lact...
A4	aband			
B4	✓	x		
C1	✓	✓	✓	<u>noted.</u>
C4	x without	✓	✓	lactant
E1	✓	✓		sip
E4	✓	x		
F1	✓	✓ part		
F4	✓	✓	✓	lact..., Malt...
O4	✓	x		
#1	✓	x		
G4	aband.	maybe ill.	→	rec. no ♀
D1	✓	✓	✓	lact...
G1	✓	✓	✓	
H4	✓	✓	✓	lactant lactant lactant no lactant

DATE: Oct 5, 1954

REF: [178]-179-180

Cross in 10: ratios W2401:W2344/4. ca 8⁴⁵ A.M. Cross is therefore somewhat old when picked (10³⁰ - 11¹⁵) = 1:45 - 2:15 hours. 16 pairs were picked initially. Results:



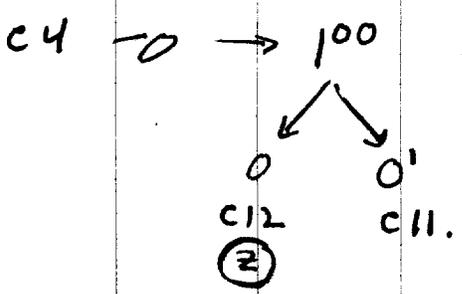
B12: Lac⁺ Ara⁻ / ♀

A2: Lac⁻ Ara⁺ / ♀

A4. ~~✱~~ abandoned to complex $\textcircled{B1}$?

B4 - 0 s.p ♀ died

C1 complex ~~✱~~ = ^{mutability} What is C3 - originally listed as motile. ~~✱~~ ^{save C56 C5 A565} no!



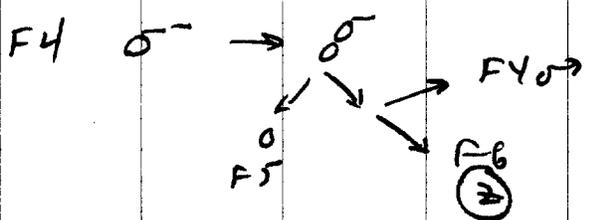
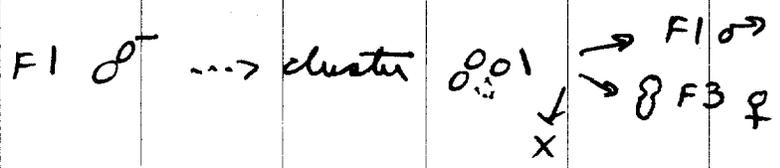
C11: male with head away.

Note C12 exception.

C12: $\frac{\text{Lac}^+ \text{Ara}^+}{\text{♀}}$ only

E1 - 0 s.p both survived \rightarrow E1 σ
 \rightarrow E2 ♀

E4 $\sigma^- \rightarrow \sigma_1$ ♀ died \rightarrow E4



Note pairing combinations

FG: $\left. \begin{matrix} \text{A} \text{♀} \\ \text{C} \text{Lac}^+ \text{Malt}^+ \text{Xyl}^+ \text{SR} \\ \text{D} \text{Lac}^+ \text{Malt}^+ \text{Xyl}^+ \text{SR} \end{matrix} \right\} \text{Ma}^- \text{Xyl}^-$

(23 + 5) Malt⁺ tested no lact⁺ Malt⁺ found

no ss

DATE:

REF:

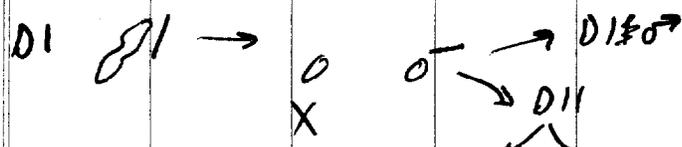
1 2 3 4 5 6 7 8 9 10

D4 ♂ all ♀ exc. eventually died, not before breeding

H1 ♂ → H1 ♂ (fully spout exp.)
 ↓
 all 6 ♀ died!

G4. conferred ♂ → G4 both ♂ ♀ died?
 ↓
 G5

For fuller pedigree see below.

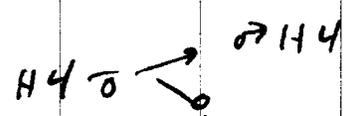
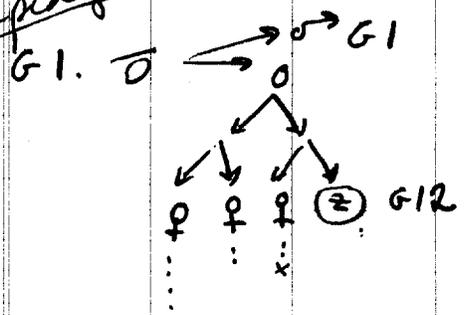


Presume correlation
 D11
 D21-22-23-24 } lact...
 D26

D11 D13 ... 7 progeny all ♀: D13, 16, 14, 25 }
 B13, 23 }
 C16 }

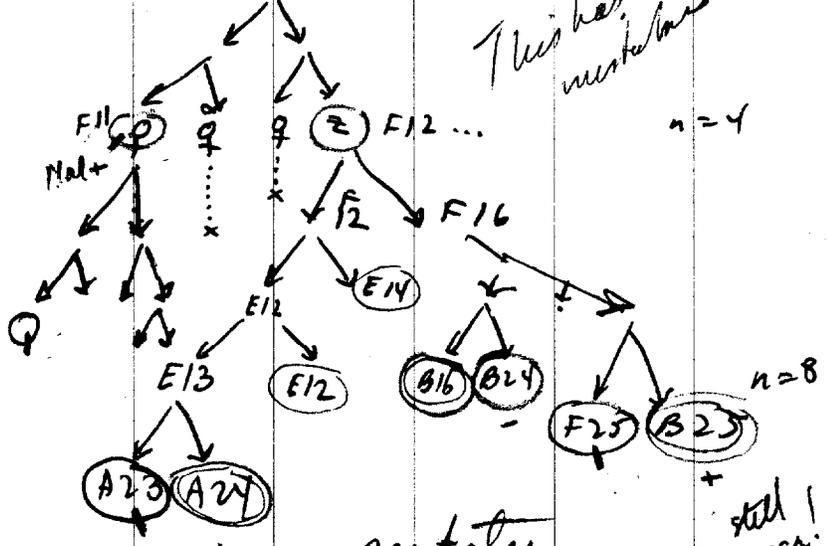
6 progeny all segregants?
 1 inviable

see pedigree



see pedigree

Thicker, mutant line.
 n=4



see later

still seg!

zygote summary:

viable excipient in
A1, C1, C4, E1, F1, F4, D1, G1, H4.

z in O = 1/9

pairings seen in C4, F4, D1,
 and probably correlated in 2/3.

DATE:

REF:

D(M)D(H) Note

	#	Lec*	Ara	Mal	Xyl	MFL	Gal	S/lac	9	10
I A.	1	A 2	-	+	-					
	2	5	-	-	-					
	3	6	-	-	-					
	4	3	-	-	-					
	5	B 2	-	-	-					
	6	C 5	-	-	-					
	7	6	-	-	-					
	8	E 2	-	-	-					
	9	F 3	-	-	-					
	10	5	-	-	-					
II	1	G 4	+	+	+	+	-	+	S	+
	2	B 12	+	-	-	-	-	+		
	3	13	-	-	-	-	-	-		
	4	C 11	-	-	-	-	-	-		
	5	12	+	+	-	-	-	+		
	6	D 11	+	+	-	-	-	+		
	7	13	-	-	-	-	-	-		
	8	16	-	-	-	-	-	-		
	9	E 14	-	-	-	-	-	-		
	10	15	-	-	-	-	-	-		
III	1	C 16	-	-	-	-	-	-		
	2	F 11	-	-	+	-	-	+	Mal - only	
	3	11	-	-	-	-	-	-		
	4	16	+	-	-	-	-	+		
	5	16	+	-	-	-	-	+		
	6	G 11	+	+	-	-	-	+		
	7	12	+	+	-	-	-	+		
	8	14	-	-	-	-	-	-		
	9	15	-	-	-	-	-	-		
	10	16	-	-	-	-	-	-		
IV	1	A 21	-	-	-	-	-	-		
	2	21	-	-	-	-	-	-		
	3	21	+	-	+	-	-	+	Mal - only S!	
	4	26	-	-	-	-	-	-		
	5	B 23	+	-	-	-	-	+		
	6	23	+	-	-	-	-	+		
	7	26	-	-	-	-	-	-		
	8	23	+	-	-	-	-	+		
	9	26	+	-	-	-	-	+		
	10	26	+	-	-	-	-	+		

I
A.

II

III

IV

wh. Lec+

* if streaked, ✓ = pure; mixture as indicated

(MFL, Mal - only)
all R exc. as noted

all on the types or sup.

---+
+-; no ++

+- only

+- only

— singular has colour.

c22A — pure Zalt
lazt. save

c3: non visible. 2 colour eyes, pure Zalt. ∴ typ. ♀.

DATE:

REF:

D(n) D(H)

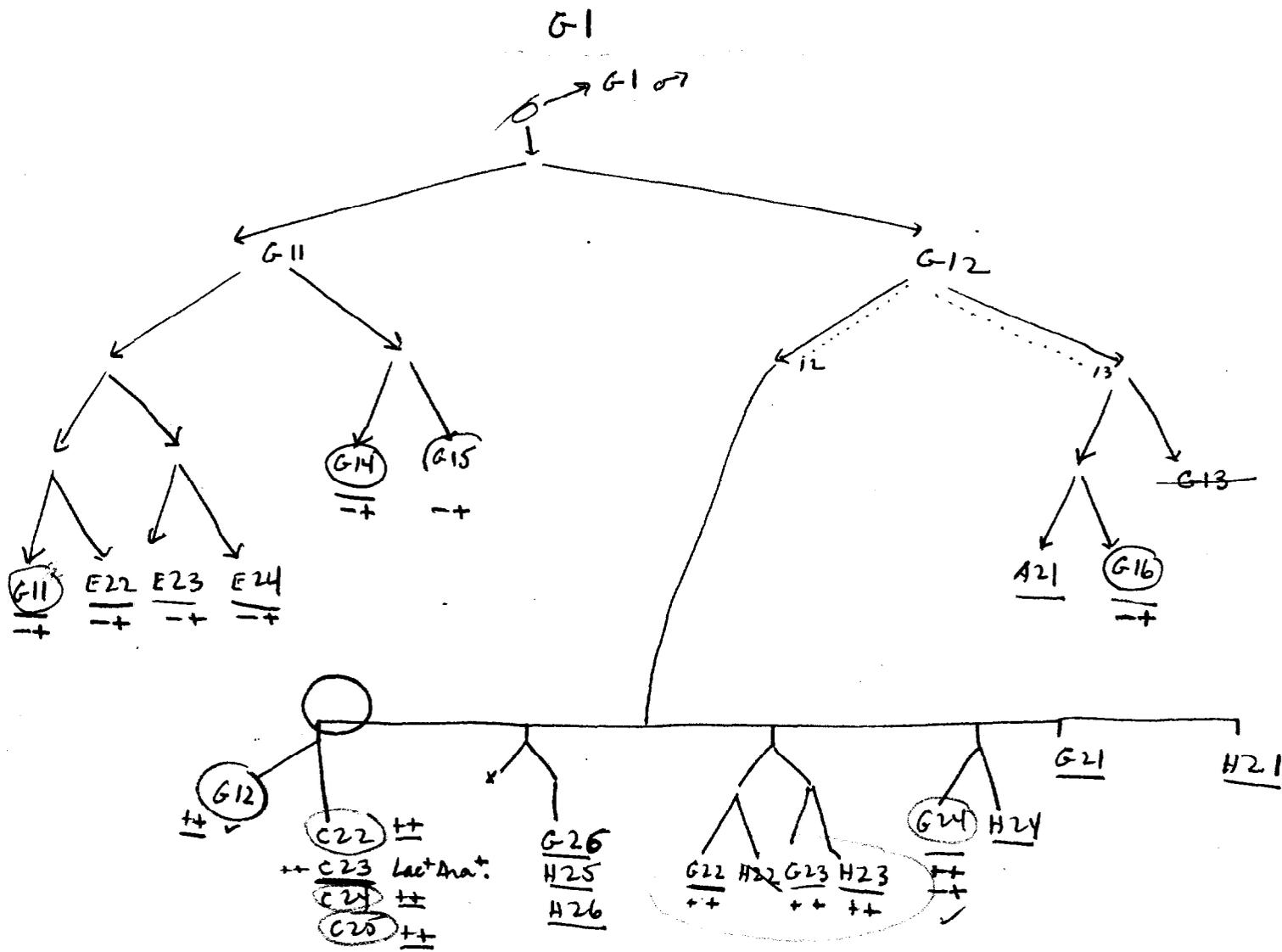
	#	Lac	Ara	Mal	Xyl	MPL	Gal	S/Pac	9	10	
V	C21	+ -	-								
	2	+ -	+								
	3	+ ✓	+								
	4	+ -	+								
	5	+ -	+								
	6	26	-	-	all -	all -	all -	all +			
B. A	7	+ ✓	-								
	8	+ ✓	-								
	9	+ ✓	-								
	10	-	-								
	11	-	-								
	12	26	+ ✓	-							
IV	1	-	-								
	2	-	+								
	3	-	+								
	4	-	+		all -						
	5	-	+								
	6	-	+								
	7	-	+								
	8	25	-	-	+ ✓	-	-	+	S/orz.		
	9										
V	1	-	+								
	2	+	+								
	3	+	+								
	4	+	+								
	5	+	+								
	6	-	+		all -						
	7	-	+								
	8	26	-	-							
	9										
	10										
VI	1	+	+		+	+	+	S			
	2	+	+		+	+	+	S			
	3	+	+		+	+	+	S			
	4	+	+		+	+	+	S			
	5	+	+		+	+	+	S			
	6	↓	↓	↓	↓	↓	↓	↓			
	7	↓	↓	↓	↓	↓	↓	↓			
	8	↓	↓	↓	↓	↓	↓	↓			
	9	↓	↓	↓	↓	↓	↓	↓			
	10	↓	↓	↓	↓	↓	↓	↓			
	11	↓	↓	↓	↓	↓	↓	↓			
VII	12	↓	↓	↓	↓	↓	↓	↓			
	13	↓	↓	↓	↓	↓	↓	↓			
	14	↓	↓	↓	↓	↓	↓	↓			
	15	↓	↓	↓	↓	↓	↓	↓			

4 S. irregular colonies A. distinct orange

no lac except S/orz.

M-H- ~~NOT TESTED~~ with yo!

see also 64



- ♀
- Lac - Ara⁺ / Ara⁻
- Lac⁺ / Lac⁻

- pure as in d
all Mal - Xyl - MR - SR

Still to be characterized:

- ① G11 purity in Ara (Lac - Ara⁺). Try V₁ also
- ② Look thoroughly for recombinants of lac/Ara in the G12 progeny. (~~Use V₁~~)
- ✓ ③ G24 any complementary Lac⁺ Ara⁻?
(No - of 45 Ara⁻, all Lac⁻; do. Lac⁺)
other three types definite = A B C
-- ++ -+

$\frac{0}{+} = \text{lac}^- \text{gal}^+ \text{ara}^- (\text{Mal}^- \text{S}^R) (\text{Xyl}^- \text{MH}^-)$

or ... + - + - s + +

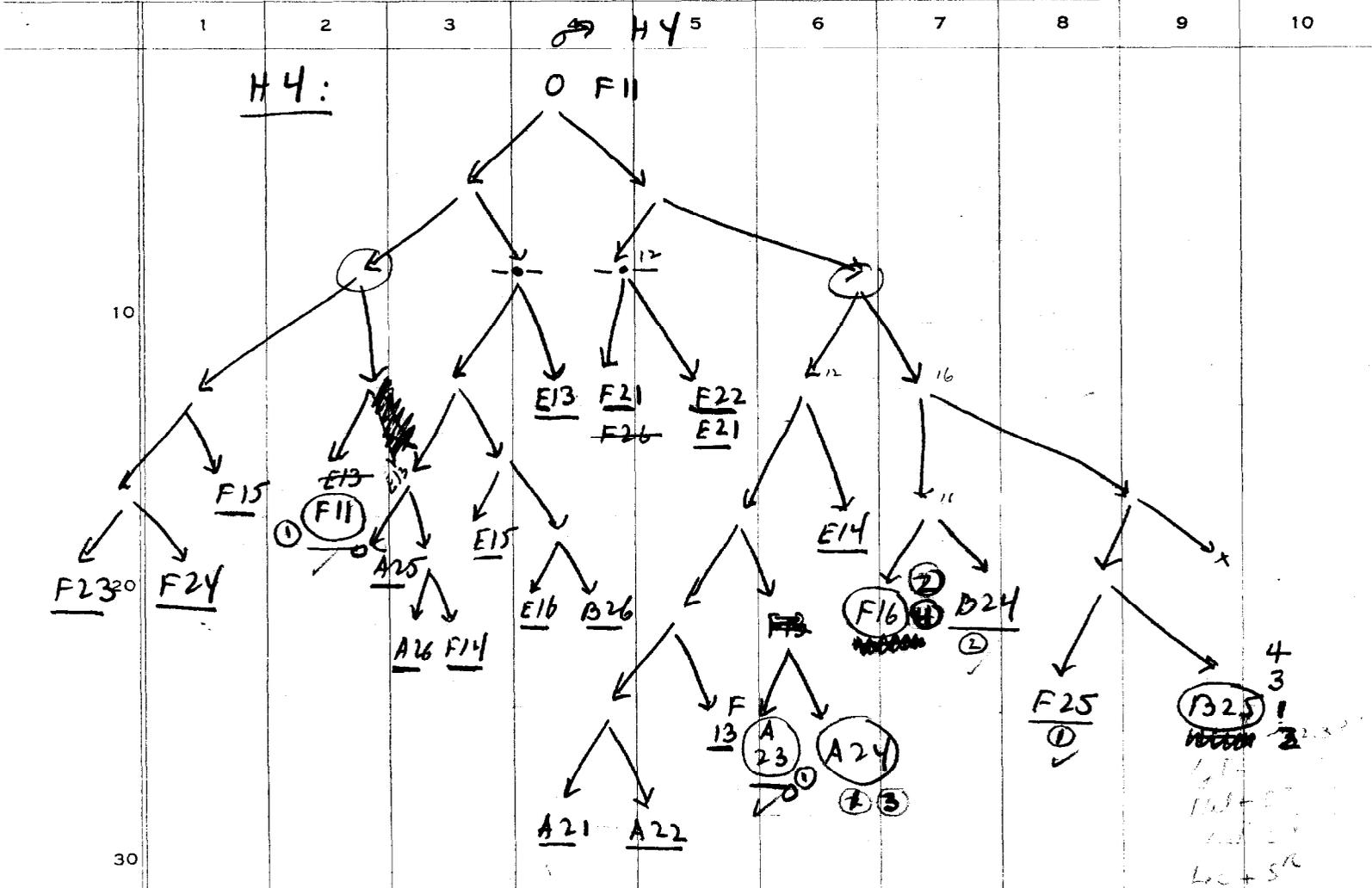
pure ara⁻ gal⁺ MH⁻

Redraw pedigree

1186

DATE:

REF:



- = Lac⁻... SR
 o verified in mixture
 - pure
 ○ segregating

① Mal⁺ S^S Lac⁻ Xyl⁺ ② Mal⁻ SR Xyl⁺ Lac⁺ ③ Mal⁺ S^S Lac⁺ Xyl⁻ ④ Mal⁻ SR Lac⁻ Xyl⁺

F25: Mal⁺ S^S Lac⁻ pure

B24: ^{pure} Lac⁺ Xyl⁺ SR Mal⁻

50 F16: "Lac⁻, + SR; Xyl⁺" Mal⁻

all are Mal⁻ Gal⁺ M⁻ H⁻ Ara⁻ pure

F11 Mal⁺; Lac⁻ SR/SR.

A23 " "

A24 Mal⁺ S^S < Lac⁺ / Lac⁻ > Xyl⁻

B25 any Xyl⁻ SR ~~SR~~ NO
Lac⁺ SR: all Xyl⁺

General details

Cauluscais

- a) heterozygous vs. heterozygote
- b) double mutants?

B25 etd. 10/14: 2 plates replicated to Lac ± Sur
Xyl
Mal

① all Lac⁺ S^R are Xyl⁺

② all S^R = Mal⁻

③ all Mal⁺ = Xyl⁻

Mal⁻ = Xyl⁺

④ 1, 2, 3, 4 types seen.

also, none of Xyl⁻ S^R, ♀ is absent.

Further tests on content of clones

1186

DATE: Oct. 11, 1974...

REF:

	1	2	3	4	5	6	7	8	9	10
C:	Lact Lact+ and - , to check F6. 10 lact Xyl 10 lac- ∴ F6 includes ✓				Mal S, Xyl. Xyl S all- R all- R		Mal all- 5+ 5-			
10					Lac+ Mal- Xyl- SR lac- Mal- Xyl- SR lac- Mal+ Xyl- SR		Not excluded: lact Mal+ (pur. Xyl-SR) S ³			
20	B25 12 lact 16 lac-				Xyl R - - ↑ medium?		S S S Mal+ all+		should be same S ^R ! repeat (might have confused F25)	
30	B12 4 + 4 -				- - - -		R R R R			
40	F16 2 + 2 -		? →		- - - -		R R R R			
40	A24 1 - 1 +				- - - - medium v.g.		S S + +		do. in fact of replicates to isolate Xyl- lac+ Mal+ S ^R lac- Mal+ S ^R	
50	C22 { lac 3+ 24 " 2- 25 " "		A24 { 3+? 3- } " " " "		C12 G12 C21		4+, 4- " " 1-+?) 4+-		∴ C22-24-25 have lac+ A24 / lac- A24 C24 has ++, -- and -+ ?	
all SR 11 lact + A24 infect side lact were not same										

all Mal+ are S^R
Mal- S^S

B25

Genotype	Type
Mal- : 5 lac- Xyl+ Xyl+ S^R	B25A (4)
Mal+ 7 Lact	B (3)
3 Lac-	C (1)
	D (2)

no Xyl- S^R on streak

any ♀?

A23 4+ Mal } all lac- Xyl- (1) A23B = c
4- " } (♀) A23A = ♀

F11 ditto } lac- Xyl-

A24 1 + 7 } Malt+ S^S Xyl- (3) A24A
1 lac- } (1) A24B

F16: A (2) - 2 (Lact) both Xyl+ ✓
B (4) - 4 (Lac-)

Types seem to include

♀	Lac ⁻	Xyl ⁻	Mal ⁻ S^R
(1)	-	-	+
(2)	+	+	-
(3)	+	-	+
(4)	-	+	-

Note complementarity

two lac classes included here!
lac+ possibly created

In synthesis of two plates

incl Lac, Lac⁺
Mal, Xyl,
only (1-4) found, no ♀, no Mal/Xyl
no S^R

Selected and unselected Hfr: fertility;
Hfr x Hfr.

1187

DATE: Oct. 8, 1954.

REF:

181

overnight cultures:

	1	2	3	4	5	6	7	8	9	10
A	11851	C1	♂	(futile)						
B		C2	♀							
C		D1	♂	(infertile)						
D		D2	♀							
E	W2582.									
M	♂ W2344M1									
F	♀ W2401									

Embryon - no gross difference
in fertility of a fertile vs. infertile
pair! also no prolonged adolescence
required for re-mating.

1:40 PM. Mix in 7ml broth 0.1 ml each of ♂'s and ♀'s.
(E+M at 1:1). 4:10 plate out on EMS lac⁺ sm.

Counts are lac⁺/total on lac⁺ sm

AB	5/293 (1.7%)			
	10/493 (2.0)			
CD	16/648 (2.5)	39/1090 (3.7)		

EM. Ca 3/1000 lac⁺ sm.

Try for pair collection

A9. Mix same (7 day) cultures 10⁸ ~~10⁸~~ 1E 10E:10⁸
945 AM

About 10 "pairs" isolated 1811.

But all proved illegitimate. However, cross was very late
(1:15), i.e., at least 3 1/2 hours.

DATE: October 12, 1954.

REF: 1182

W2582 x W2344 M1.

9:30 - 11 AM vic.

Most pairs illegitimate. 3 Legit, 1 shows recombination (W2582 as ♀); F1 indeterminate because mixed.

	1	2	3	4	5	6	7	8	9	10
			lac ✓	Gal	Mal	Xyl	Swarm.	Pro		NOTE
10	A1	→	±	-	+	+	+	+	S	S illeg.
X	B1	•	++ mix	+	+	+	+	+	S	+ Not ally.
	C1	→	± ✓	-	+	+	+	+	S	S illeg.
	D1	→	± ✓	-	+	+	+	+	S	T
	D2	→	- ✓	+	-	-	-	-	S	Legit
	E1	→ ♀	- ✓	+	-	-	-	-	R	x
E3	E2		+ mix	+	+	+	+ (-)	+	+	Recomb?
	F1		++ mix	+	+	+	+	+	+	
X	F2		- ✓	+	-	-	-	-	R	
	F3		- ✓	+	-	-	-	-	R	
	G2A		- ✓	+	-	-	-	-	R	
	G3		+ ✓	+	-	-	-	-	R	
	G4		± ✓	+	+	+	+	+	R	Vact)
	H1		± ✓	+	-	-	-	-	R	same as?
X	H2		+	+	+	+	+	+	R	1 +, - mixed how?
30			mixed ±, ++							

parents are W2344 = Gal-lac+ + ... " ♂"
 W2582 = Gal+ Lac- ... " ♀"

Interest: G3 = Lac+ Gal+ Pro+ Mal-Xyl-
 (18 (G3 = W2401))

may need a recombination for motility of W2344

Restrict lac and Gal ≠ B1, E2, F1, G3, H2

B1 - "both motile"

E2 - "x"

F1 1/0

1. (G3?)
 00 F2

G3 } 10 → 0
 G4 } 10 → 1 → ♂

flush partner.

m lac

may be recomb in opposite sense?

50

F1 may represent

~~for~~ W2344 x W2582 ♂

Probably not.
a mixture of
Gal+Lac - and
Gal-Lac+
mostly unpaired

G3 is evidently W2582 x W2344 ♂

B1 Lac Gal
mostly ± mostly -, few +

E2 ± and - - and +
? are all Lac - Gal+?

F1: Gal+Lac - pool
all non mutated

F1 mostly all ± mostly -

M2 " " "

since F1 is mostly
Lac-Gal+, it is
presumably ♀ + few ♂

G3 pure +

pure + ; legs on tra

G4 all ±

~~all +~~ not tested

res. -

• terminate
• capture

♂ x ♀ to
polymer

1189

OCT 14 1954

DATE:

REF:

183

	A	B	C	D	E	F	G	H	9	10
1										
2										
3										
4										
5										
6										

gww:?	1	2	3	4	5	6	7	8	9	10
183	0, d	0, d	0, d	+ nm	+ m	0	+	0		
183	0, d	+ d	0	+ m	0	+ m	0	+		
183	0	+ nm	0	0	0	0	0	0		
183	not	0	+ m	0	0	0	0	+ sparse		
183	0		0	0	0	+ nm	0	+ nm		
183			0			0	0			

abandoned

why poor or no growth

① low temperature

② capillary tip too sharp? or too acutely bent

but same as 190, 191

Abandoned owing to growth failure (superstructure?)

1190 Sws.

	→	♀	zyg.	types.
B4	✓	✓	no	
C4	✓	✓ part	—	
	✓	✓	0	

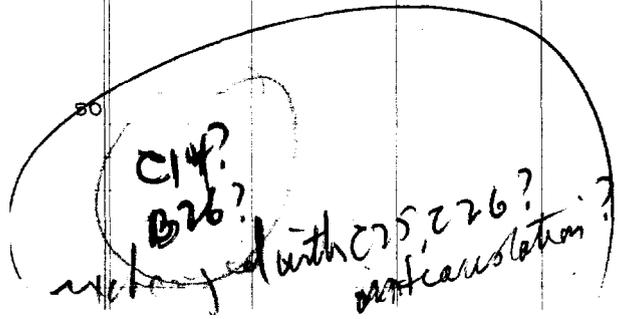
DATE: Oct 19, 1954

REF: [185-186-187]

	1	2	3	4	5	6	7	8	9	10
	W-2344M x W2401		10:30-12N	10;1 Q		Collect numerous pairs and follow for				
	1-3 exconjugant generations. to [186-187]				P19: refrigerate [185] after transferring three pedigrees B4, C4, E4					
				1	2	3	4	5		
10										E L
					B5	A15	A15	A15 ✓ A24 ✓		
						A16	B15 ✓ A16 ✓ A22 ✓	B13 ✓ B16 ✓ B14 ✓		
				B5		A16				
20					A2	A13	A13 ✓ A14 ✓ A11 ✓ A12 ✓	A13 ✓ A25 ✓ A14 ✓ A23 ✓ A11 ✓ A26 ✓ A12 ✓ A21 ✓		
					<					
30					C11	C11	C11 ✓ D11 X D23 ✓ C12 ✓ C26 ✓ C13 ✓	C11 ✓ D12 ✓ 18 D11 X D23 ✓ 19 C12 ✓ 20 C26 ✓ 21		
				B6						
						C14	✓ v.s. C14 D14 ✓	D14 ✓ 23 D22 ✓ 24 C15 ✓ 25 C25 ✓ 26 D15 ✓ 27 D13 ✓ 29		
40						C15				
						C15				



These 3 pedigrees: all q on lactal Ha Mal Mtl sm.



29 alls

DATE:

REF:

	1	2	3	4	5	6	7	8	9	10
		1	2	3	4	5				
10							EM			
20			E15.0X							
				E13	✓	E13 ✓ E22 ✓ B26 ✓ E26 ✓ E14 ✗ F21 ✓ F15 ✓ F23 ✓	1 2 3 4 5 6 7			
				E14	✓					
					F15					
30				E11	✓	E11 ✓ E23 ✓ E25 {stomachy} ✓ ✓ ✓ E21 ✓	8 9 10 11 12			
				E12	✓					
					E24	← ←				
				C16	✓					
					D16	D16 ✓ D24 ✗ E16 ✗ F24 ✓	17 18 19			
40				E16	✓					
					F16	✗				
50										

24.5 → ✓
25 ✓
25 ↓

C6

E24 B23
B24 B25

Sp. 1
Xdidnot

DATE:

REF:

	1	2	3	4	5	6	7	8	9	10							
10																	
20				E5			G11	G11 x									
								G24 ✓	1								
							H11	H11 ✓	2								
								H25 ✓	3								
							G12	G12 x									
								H12 x									
							G13	G13 ✓		4							
								H13 ✓		5							
							G14	G14 ✓		6							
								H14 ✓		7							
							30				E6			F11	F11 ✓	8	
															F25 ✓	9	
G16	G16 ✓	10															
	G25 ✓	11															
F12	? NF		G15 ✓														
	G15 ✓		G15 ✓														
			G26 ✓														
			G26 ✓														
F13	F13 ✓		F13 ✓														
	F26 ✓		F26 ✓														
40							H16	H16 ✓	12								
								H26 ✓	13								
							F14	F14 x									
								H15 x									
50																	

AK

E4 → E5, 6

DATE: Oct 22, 1954

REF:

1 By UV/EMB, best obtained 43 lac- mutants from W2654.
 A is nearly full- ; B + C are slow but entirely scoreable
 P21 (no D(0)) : W2654, ~~W2654~~, W2663. Keep A as W2663

10 215 P22 Prepare mixture of W2654 + W2663 1:1
 (no D(0)) with .01 and .001 ml / 10 of each & mixture. 37°

	Tube	1	2	3	4	5	6	7	8	9	10
A	W2654	.01	1-3								
B	"	.001	1-3								
C	W2663	.01	1-3								
D	"	.001	1-3								
E	Mix	.01	1-5								
F	"	.001	1-5								

A1 became + 11/7 AM. A2 ± 11/8.
 → almost pure lac- mutants.

11/8: 1:± 2:± 3:0
 → almost pure lac- mutants

also tube #0 = D(0).

P23 (out in bench all day). Streak on EMB lac - O-tubes and DA tubes. Turbidity 0 except in D(0).

A-D (0) as parent pure E ca 1:1 F > 20:1 lac- : lac+

40 F~~1~~: (DA washmore) ca 1:1
 E, F1 (Ara) ca 1:1
 P24 0 turbidity.

P9 streaks of E1, E2
 E1 now + E2 ± E3 0 E4 ±
 F1 now + B1 + B2 ± B3 ±
 F2 ± A2, 3 ±

structured P9:

E1 }
2 } almost pure +
3 }
4 }

F1 }
2 } almost pure +
3 }
4 }

A1 ++
C1 --

E0 almost pure +

F0 almost pure +

clear that "baz -" does not present in S^R some
(of S^R !)

DATE: 10/23/04

see 1190

REF:

188-189

	1	2	3	4	5	6	7	8	9	10
	A	B	C	D	E	F	G	H		
11	✓ ⊙	✓ ⊙	x ⊙	⊙	⊙	⊙	x ⊙	x ⊙		
12	x ⊙	x ⊙	x ⊙	x ⊙	x ⊙	⊙	x ⊙	x ⊙		
13	✓ ⊙	⊙	x ⊙	x ⊙	⊙	xx ⊙	x ⊙	⊙		
14	✓ ⊙	x ⊙	x ⊙	x ⊙	⊙	x ⊙	⊙	⊙		
15	✓ ⊙	x ⊙	x ⊙	x ⊙	⊙	x ⊙	x ⊙	⊙		
16	✓ ⊙	⊙	x ⊙	x ⊙	⊙	x ⊙	x ⊙	⊙		
21								x		
22								x		
23								⊙		
24								x ⊙		
25								⊙		
26		⊙		⊙				⊙		

188

189

A24. - regrowth
x 11.9.

Picks 188: A 11¹ B 11⁷ D 11² E 11¹³ F 11¹⁵ G 11¹⁸ H 11²⁰ B26²⁴
 13² 13⁷ 14² 12¹⁵ 15¹ 14²¹ D26²¹
 14³ 16² 16¹ 14⁷ 16² H23¹
 15¹¹ 16¹² 16²³ 25¹
 16⁵ 26¹

also viable: ♂: A4, B4, C1, C4, C4, F1, G1, H1, H4 second
 ♀: A6, B6, D2, D5, D6, F first

copy 40
 photo
 put in cell

A26 { all ♀ except G15 are lact -
 all ♂ lact ±.
 G15 pure? lact

No good pedigrees after 3 day, upr. storage

over.

A27

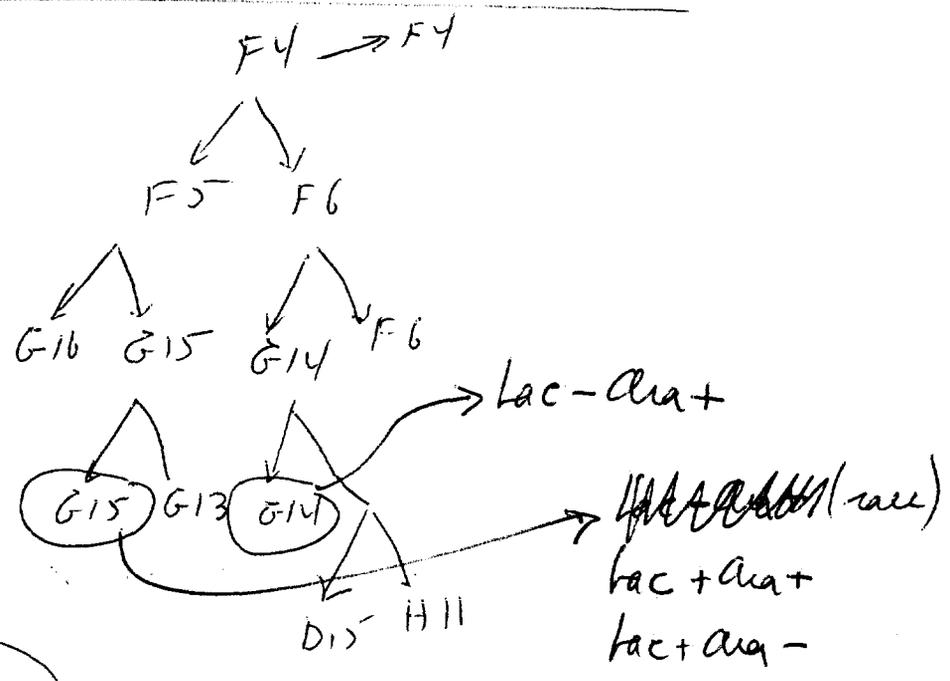
all viable clones from (185, 188, 189)

are parental on Lac, Gal, Xyl, MTH, Ara, Gal sup.

except # 19 = Lac⁺ s^x Ara⁺ } Gal⁺ MKM⁻
18 = ara⁺ lac⁻ }

85-1-2 should be selected on EMS Gal for purity.

save
AG
156



all but G14 G15
inviable

G14: pure ara⁺ Gal⁺ Lac⁻

G15 ara⁺|- "

For future tests, use "MKM" (Malt + Xyl + MTH) EMS in preliminary screening.

also

ca 7/14-16

look at Caulobacter.

Island from Hutner

is contaminated with

interesting metal rod

($\frac{1}{2}$) ② grows better

~~than~~ in presence

than NSB, and OK

at 37°

26-2a FEB 26 1955 Photo
26-2b
27B4 (small plate, range #)

27B1
27B2
27B3

29B2
29B1

FEB 26 1955

28D



should be 28D1.

DATE:

REF:

1 2 3 4 5 6 7 8 9 10

G14: pure lac - Gal+ Ara+

G15: pure Gal+

Reported rare lac - among many Lac+.

of 12 Ara+ all Lac+

12 Ara- all Lac+.

10

at least 1 lac - Ara+ identified.

Reexamine for

lac - Ara-. V_1 should also be scored: both V_1 R m Lac.

20

An restreak, G15 was pure Lac+ (Purified Lac - colony probably came from G14.)

Absence of lac - Ara- is notable

but might be in other parts of pedigree, undetected.

If sequence is

lac - V_1 - Ara, implies crossover of V_1 - Ara. Ara - should be checked also.

Ara - should

30

late V_1 and restreaks! - These are all Lac+ V_1

Some lac? and cause of

Ara - are Lac+ V_1 .

Types recovered are

Lac+ Ara+ V_1

Lac+ Ara- V_1

and Lac - Ara+ V_1 as if

3/4 strands from

$\frac{+ + r}{- - s}$

hybrid with r.o. between Lac/Hcr. Unfortunately that others were not recovered.

40

Note A5 and A6 also had mottled appearance in EM33al, but DC failed to find any evidence of segregation.

50

Sequential, if one r.o. is recovered, should there be also?

to periodic selection coming in?

Reconstructed by Luca

stage 14-5

11/27 Pick 109 singles from EMB-0
also 8 clusters of 5, to EMB-lacI sm.

None of singles, 2 of 5's showed S^R

Restreak these on EMB-lac for final eval.
= total of 2/149. A, B.



W2716

10 colonies on EMB-lac. Pick at random
3/10, 2/10 S^R

Parallel platings ^{at least stage} showed $S^R = Gal^-$
(if S^R suppresses S^{gal+}). This isolate has weak if any
response to S^{14} .

Note C Gal^+ and \pm ? together noted as

~~C1 = $S^R Gal^-$ C3 = separate isol S^S~~
~~C2 = $S^S Gal^+$ Gal^- - - not on second hand, probably matter of fading~~

Galt reversions of W2716 were noticed to be Gal⁺!

passed through 20 passages (ca 10^{-3} ml/10 - 10^{-4} ml), i.e. for

1/10/55 about $\log_2 10^{80} =$ about 250 generations, then plated.

DCC examined ca 1000 colonies (5 plates); all were SR on replica. One isolated as W2716-20 for quantitative comparison.

about 0.1% of colonies at this stage were Gal⁺. Proved still SR and, as above, unstable +. Same ①.

Tz and crosses

1195 ¹¹⁷
~~1194~~

DATE: OCT 27 1954

REF:

1 2 3 4 5 6 *Smegmatolimus* 8 9 10

Grow ♂, ♀ overnight. (1194) not recorded!

8:55 add 1ml broth culture to .1ml .05% Tz

10:05 - treatment 2: spin down + resuspend
 " 3: use stained cells for use

ca 10²⁵

- ① ♀ + ♂ 3 1ml: .5 7ml necessary
- ② ♀ + ♂ 2 " " "
- ③ ♀ 2 + ♂ 1ml: .5 " 37°
- ④ ♀ 3 + ♂ " " "

Also note .2ml Tz went slightly faster.
 (2ml Tz)
 addition of 1ml fresh broth at 8:55 delayed coloration about 1 hour
 ♂ reduced Tz > ♀.

Exp. n.g. - label (Tz) insufficient for low power determination.

Conclusions - so far, Tz label has not been satisfactory.
 In growth overnight in Tz, much of the label is extracellular. In higher periods so far, there has generally been just too little label to be valuable. Needed: some pulvis. system incorporating the label, especially in line 28. This should not be allowed to interfere with pulvis work and cytology.

DATE: Nov. 2 1954

REF: 192

11/1 Prepare ♂, ♀ T₂ for peel. study
General conclusion. (T₂ label can be introduced in
ca 2 hours (in old both .005% T₂)

10 (T₂ diffuses considerably with motility
but some pairs may still be obtainable.

P1 prepare labelled cells. let stand in fry:

20 A2 Most ♂ T₂ had celled. Supernatant may contain the
motactive labelled ♂♂. Take off about .4 ml and mix
with ♀ unlabelled + ca 1 ml both 10:30 AM.

(also prepare freshly labelled ♀♀ and fish ♂♂

30 General, only a few labelled ♀ proved satisfactory.

40

50

DATE:

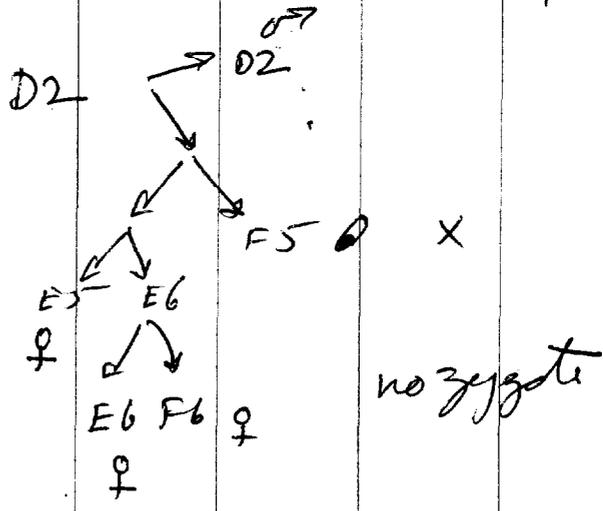
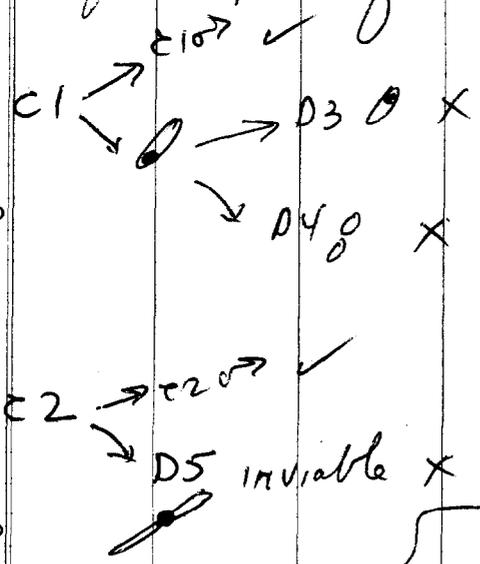
REF:

192-193

Comment: 192
 E1b, c, are ♀ ♂ →
 C2, C5 are ♂ →
 F2, F3

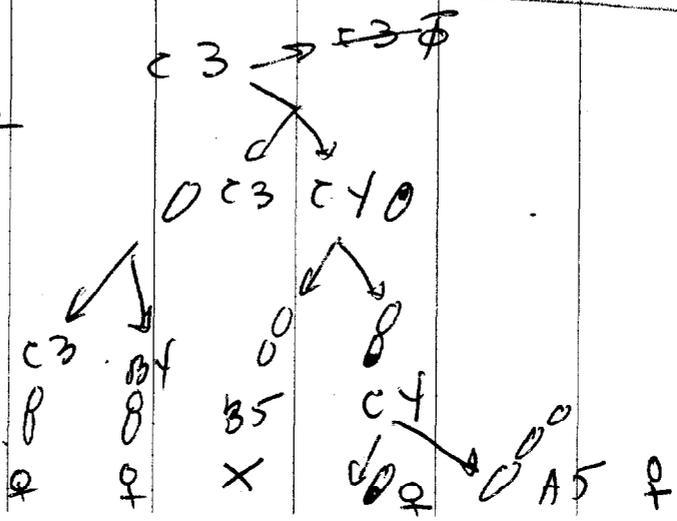
~~192~~ 10 C5 was given as non motile unless examined. Not examined in detail; may have been sterile pair. (Examined now)
 C5 OK. But examine for motility of present culture.
 F2 - F3 no record!

20 192 was attempt to follow pedigree & T2 under the following are of interest: Most ♂ did not grow



no zygote

30
 40
 ♂ survived at least 3 generations but no zygotes.



50

DATE:

REF:

	1	2	Lac	Gal	His	M ⁺ M	S ⁺ M	8	9	10
1	C1		+	-	+	+				
	D2		+	-	+	+				
	E1a		I	-	+	+				
→	92 E1b		I	+	-	-	R Lac-			
→	E1c		-	+	-	-	R Lac-			
	193A1		+	(+)	+					
	2			error?	+					
	3				+					
	Y				+					
10	5				+					
	Ab				+					
	C1									
	C2									
	C4									
	C6									
	E2									
	3									
	4									
	5									
20	6									
	G1									
	2									
	3									
	193H2									
25	192 E2		+	+	+		R - +			
							R - +			
31	192 A2		-	+	-	-	R -			
	3		-	+	-	-	R -			
	5		-	+	-	-	R -			
	192 B4		-	+	-	-	R -			
→	39 192 C2		+	-	+	+	S -			
→	36		-	+	-	-	R -			
→	37		-	+	-	-	R -			
	192 C5		+	-	+	+	S -			
	E5		-	+	-	-	R -			
	E6		-	+	-	-	R -			
40										
N.R.	41 192 F2		+	-	+	+	S -			
	2 192 F3		+	-	+	+	S -			
	3 F4		-	+	-	-	R -			
	4 F6		-	+	-	-				
40	193 B1		-	+	-	-				
	2		-	+	-	-				
	5		-	+	-	-				
	6		-	+	-	-				
	7		-	+	-	-				
	8		-	+	-	-				
	9		-	+	-	-				
50	C3		-	+	-	-				
	C5		-	+	-	-				
51	D3		+	+	-	-	R + -			mm
	4		-		-	-	R -			
	5		-		-	-				
	6		-		-	-				
59	F2		-		-	-				
	3		-		-	-				
	F4		-		+	-	R -			mm
	5		-		-	-				
	6		-		-	-				
80	H1		-		-	-				
91-62	H4-H5		+		-	-	R - / R -			mm

97

all OK

~~Res~~

DATE:

REF:

Conclude: It has no particular value and tends to impair viability as well as motility?

[192] ER needs review! Mated as mixture! Only part may have been picked up!

[193] H2 random pairs were picked & pedigree analysis.

H2 mixed as recorded! What are H4-H5? prob A6

Pairs completed are

Total score then is:

20

♂ ♀ ♀
A2 ✓ B1 ✓ B2 ✓

A109?

A3 ✓ B3 X

A4 ✓ B4 X

A5 ✓ B5 ✓

A6 ✓ B6 ✓ H6? ✓

30

C1 ✓ D1 X

C2 ✓ D2 X

A1 # C3 ♀ D3 (R) # H5 may have B5? 5 4

C4 ✓ D4 ✓ ~~H4~~

E5 ✓ E5 ✓

40

E6 ✓ D5 ✓ D6 ✓

E2 ✓ F2 ✓

E3 ✓ F3 ✓

E4 ✓ F4 (R)

E5 ✓ ~~E5~~ illegit ♂

E6 ✓ F5 ✓ F6 ✓

50

mixed G2 ✓ G1 ✓ H1 ✓
G3 ✓ ~~G3~~ → H2 mixed: ~~H2~~ (R)

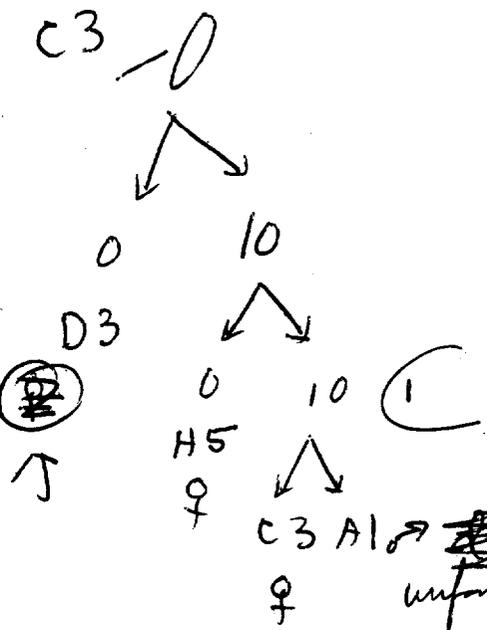
3 (B) from 14 reasonable pairs! Why so low? Pedigree analyses have indicated a higher incidence! Maybe random selection for clonal integrity in the pedigrees!

14 viable pairs
4 inviable ♀
no pedigree

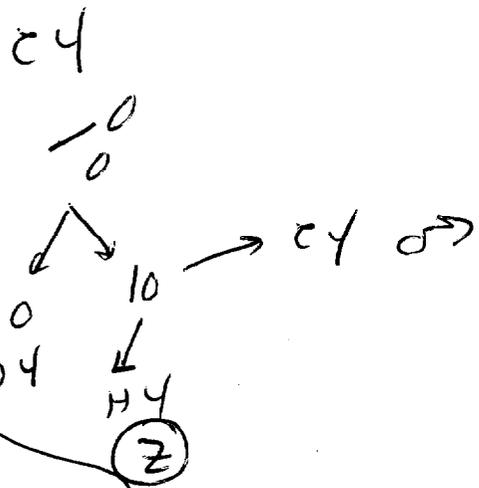
owing to temporary mistaking of one protocol sheet, not all pedigrees were clearly stated and some subs thrown out. See over.

5
6
7
8
9
10
11
12
13
14

A1-23-D3-H5
 24-04-H4
 E4-F4



~~un~~
 unfortunately not kept



prg.
 rule

exc. to prg. rule! (if rule?)

DATE: 11/7/54.

REF:

Productive pairs were

C3-D3 - Stage 10 → O D3

C3 (pres! Fate of male?)

E4-F4 10, 9, 8, 7, 6, 5, 4, 3, 2, 1 E4 OF 4.

G3-H4 10 : (10) G3 O H4

There is no ambiguity in the history of these, though it is unclear why H4 should have been chosen for G3, unless preempted.

Note reversal of pairing relationships in C3.

(cross was ~~performed~~ ^{carried} rather late)

Recombinants in σ^{σ} conjugants would be difficult to detect. Should routinely restreak σ^{σ} on EM13 lac.

Save σ^{σ} above and σ^{σ} for recheck

C3 ♂ D3 Lac⁺ - M x M - Ara⁻ S^R.

Score $V, R / S \rightarrow V, S$

E4 ♂ F4 Ara⁺ - lac⁻ M x M - S^R

$\sigma V, S \rightarrow V, R$

G3 ♂ H4 Lac⁺ - M x M - Ara⁻ S^R.

$\sigma V, S \rightarrow V, R$

See. not of very great interest.

192 E2 - mixed! But no record of separation of clones.

DATE: Nov. 4, 1954.

REF: [193]

1 2 3 4 5 6 7 8 9 10
 1:10 2:9 2A/2¹⁵-12¹⁵ setup. Ca. 24 pairs isolated and
 :7ml
 allowed to separate. Minimum of pedigree analysis except in re with -
 current fissures during configuration. Pick viruses with [192] for
 tests.

10 ~~24~~ pairs isolated on.

	17	sur.	fiss.	zyg.	type
11/51	10	✓	✓	no	
	0	x	✓	no	
20	3	✓	✓	✓	
	4	✓	x	-	

30

40

50

DATE: Nov 8, 1954

REF: Jacob letter, no. 4
CRAS culture

21079

	1	2	3	4	5	6	7	8	9	10	
	Whom cultures as month					1ml η	2ml η	7ml η	essay		
	A W1603 (W1177 hp^s)					Inc. 37° 8 ²⁰ - 10 ¹⁰ PM. Plate ca 10 ³ / EM13 bac con.					
	B W1177										
	C W1895M1 (10^2)										
	D W2344M1 (η)										
10	E W2401 (η)										
	F W2578 (W1607 hp^+)										
	Est SR+ / SR-										
	AC	54/1000				Note no marked difference in efficiency of combinations of Lp^+ , Lp^s Hfr! Try Hayes Hfr!					
	AD	37/1000									
	BC	22/300									
	BD	28/300									
	EC	10/300									
	ED	8/300									
	FC										
20	FD	too weak to count.									
30											

also get
W2588 = η
 Lp^+ η
as SR

A10 Repeat with W2344M1 (-G)
1:1:7ml 11¹⁵ AM - 4²⁰ PM. old cultures
Refrigerate to 3 PM.

	1663	1177	W1895	40	1895	2344	SR+ / SR-	Plaque/578	Results:
	A	B	E	C	D	G	AC 17/300	ca 50	measured of W2344M1 and λ^s x W2401
							AD 14/300		
							AG 3/300	ca 20	
							BC 15/200	+++ (10^2 - 10^3)	
							BD 14/150		
							BG 7/200	ca 100.	
							EC 10/300		
							ED 6/300		
							EG 0/300+ ←		

Note hp^+ x hp^+ gave more
 λ than hp^+ x hp^s . In
all combinations W1895 was
more fertile than W

also plate AC and AG, BC, BG on λ^s indicator.
(over)
(2401)

Repeat P15

use old cultures as
mostly 1:1; 7 ml
2 1/2 hours.

stretchout E1403 lac sus. Score SRT

W 2324 Motelyid	x W1177	0
"	x W2324 W2401	0
W 2324	x W1177	++ (>10%)
"	x W2401	++ also note plugging!
W 2344	x W2401	++

again P17 (A) old cultures 7:30 - 9 PM

1:1:7 (B) fresh cultures (from above) 9 PM - 10 PM

ca 1% in all B. In A, ~~W2324~~ ^{W2324} was ca 1/10% SRT + ♀

but x W1177 and ♂ x ♀ gave ca 1%. ∴ W2324 is more affected by aging than is W2344.

(C) A18. overnight cultures 1:1:7 9⁵⁰ - 11³⁰

♂ x ♀ ca 1%

♂ x 1177 > 1%

W2324 x ♀ > 1%

W2324 x 1177 +, < 1% some plugging again.
What is this phase which acts on

W2401?

Then why no of spores from pairs?

Conclusion: W2324 may be slightly less fertile than W2344. No clear evidence here of extra induction. Should use Jacob's medium, count inf.

Ectoc induction: preliminary

1198

DATE: 11/19/74.

REF:

1	2	3	4	5	6	7	8	9	10
Motility w2324 for crossing purposes - 1 passage. Too cross culture was almost sterile S.T. gave no SR+ x W1177. Zouhde probable F- (<u>w2696</u>)									

P20 10 Hour 1:1:7	W2693	x ♀	SR+ 1 papilla only in thick tract!	Believe for fertility!
	W2324	♀	ca 0.2%	
	"	W1177	2%	
	2344M1	♀	2%	

20

30

40

50

W2324 (Hayes Hfr) x W2401
single cells

1199

DATE:

Nov 11 1954

REF:

194

	1	2	3	4	5	6	7	8	9	10
	cross ① 9:40-10:30 then same time with T									
	② 11:15-11:35.									
	A1. Unseparated pair → ♀♀ only.									
	A1-6									
	A1-2									
10	B3 - A3 - B4									
	C1 - C2 - C5									
	C3 - C4									
	D1 - D2									
20	D3 - D4									
	E1 - E2									
	E3 - E4									
	F1 - F2									
	F3 - F4									
	G1 - G2									
	G3 - G4									
40	H1 - H2 - H3									
	- H3 - G4 - H4 - H6									
	Complete pairs: 9									
	Pro (♀OK) :									
50	no zygotes									

No lysis seen!
Numerous pairs despite
indifferent motility of
W2324. Cells of latter
are shorter than W2344, &
harder to distinguish from
W2401.

all parental ac factors,
14 X 14. (all+).
~~except~~ except B3 which is
lac⁺1- (lac-SK) pres.
mixed. ...
lac- : A1, A6, B2, B6, C1, C2
C4, D2, D4, E2, E4, F2, F4
G2, H4, H6.

fact: D3, F1, A3, B4, B5
C3, C5, D1, F3, G1, G3, G4,
H1, H2, H3.

Non-motiles in F1? (misc. segs)

presumed part
mixed (misc chain)
B3 - A3 - B4
Save F1, C1-C2-C5, D1-D2
as examples.

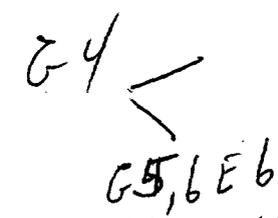
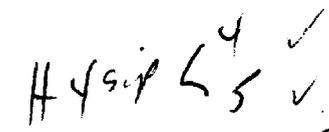
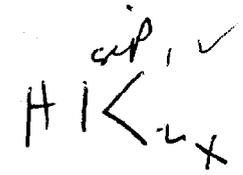
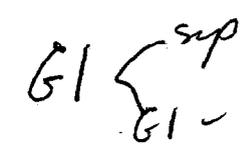
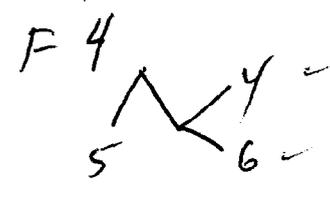
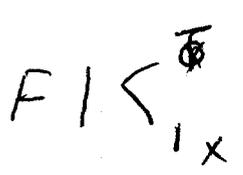
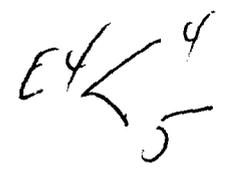
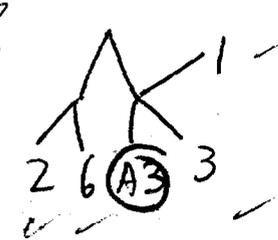
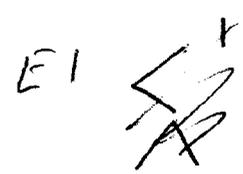
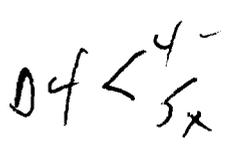
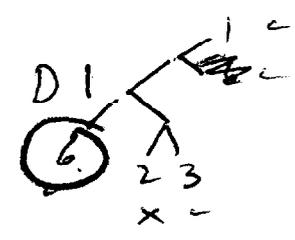
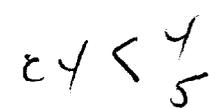
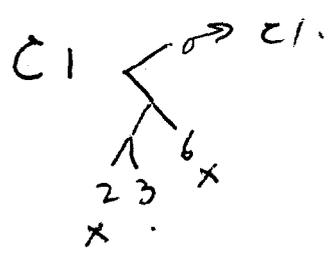
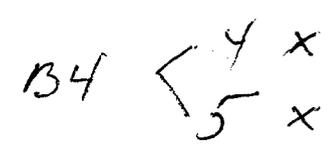
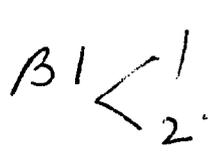
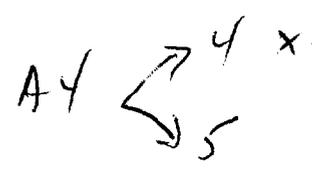
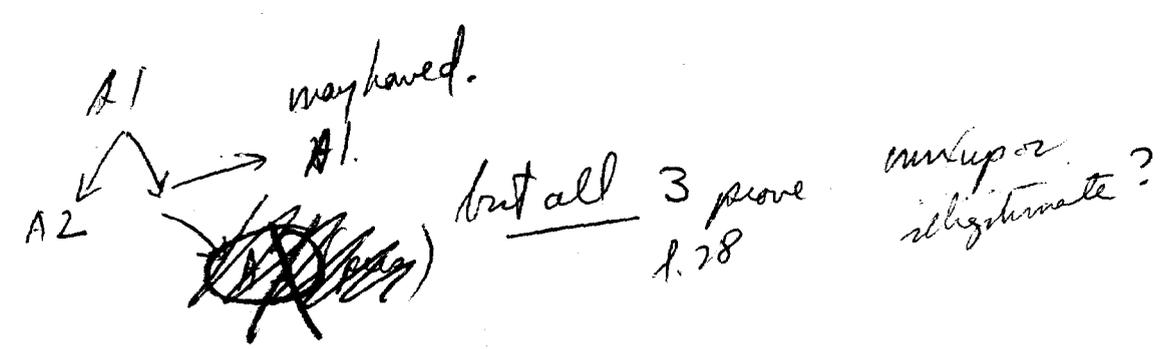
slight cross ~~W1895M1~~ x ♀
 W1895M1
 ♂

1200

DATE: Nov 13, 1954

REF: 191

	1	2	3	4	5	6	7	8	9	10
♀ D1		-10 →	① D1	0 0 2 3	① 6	Presume.		D1		
10 A3 E3 3 E6	E1	-0 →	① OE2 OE6	10 → 0 →	E1. OE3 ①A3		D2 X	3	①D6	all these lact- V ^s
20										
30										parental ♂, ♀ resp except A3 lact- and D6 lact- BU 4.9. all parental on M X M A at
40										Presumed to have been W234YM1 x W2401. However, all ♂♂ were lact+ and at least D1, E1 were T1 ^s . must have been W1895M1 instead, ♂ which had been set up concurrently! Confirm on ♂ D10).
50										Yield 1/9 c. 2/11 3490/10 1/2 Inc)



♀ complete: A5, B2, C5, (E 2, 3, 6) E5, (F5, 6), G1, H4, (sup)
 ♀ partial: (C3), (D2, 3, 6), (2), (G5, 6, E6), (9)

∴ 2/11 zygotes. save E1-2-3-6-A3 and D1-3-6